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Herring Rearing—II
The Effect of Temperature and other Factors on Development

By J. H. S. BLAXTER, M.A.

EDINBURGH: HER MAJESTY'S STATIONERY OFFICE

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CONTENTS

	Page
INTRODUCTION	3
METHODS	3
General	3
Apparatus	4
Environmental Conditions	4
Treatment of Material	6
RESULTS	6
The Effect of Egg Density on Development	6
Time to Hatching	8
(1) Estimation of biological zero or threshold temperature	9
(2) Effect of temperature on rate of development	11
The Effect of Temperature on Survival	12
The Effect of Temperature on Size, Weight and Stage of Development	15
(1) Length	15
(2) Weight	16
(3) Other characters	16
The Effect of Light, Sterility and Water Movement	17
CONCLUSIONS	17
ACKNOWLEDGEMENTS	17
SUMMARY	18
REFERENCES	18

THE EFFECT OF TEMPERATURE AND OTHER FACTORS ON DEVELOPMENT

INTRODUCTION

It is well known that temperature and other environmental factors may affect the velocity of biological processes. High temperatures will accelerate, and low temperatures retard, the speed of development of teleost embryos. This has been shown for instance by Dannevig (1895), Reibisch (quoted by Williamson, 1909), Johansen and Krogh (1914), Seno *et al* (1926), Nakai (1927), Gray (1928), Kajiyama (1929), Battle (1930) and Merrimen (1935).

Seno *et al* and Battle also found that salinity extremes reduced percentage hatching while Kajiyama found the rate of development independent of salinity within certain limits. Johansen and Krogh found that low oxygen tensions retarded development and that light intensity seemed to have no effect while Hata (1949) observed no consistent effect of wavelength on hatching. Hata (1929) found that vibration killed fish embryos.

The embryology of the Atlantic herring (*Clupea harengus* L.) was first studied in detail by Kupffer and Meyer (quoted by Brook, 1885-6). Meyer (1878), Williamson (1909, 1911) and Soleim (1942) found that the time to hatching was increased by low, and decreased by high, temperatures of incubation. Ryder (1882) observed this also in the shad (*Alosa sapidissima*). Soleim (1942) also hatched herring in salinities from 35·57‰ to 22·17‰, finding little or no difference in survival, while Ford (1929), succeeded in hatching herring eggs in water of salinity 5‰. Kelley (1946) working on the Pacific herring (*Clupea pallasii*) found that development was retarded and larvae deformed by pH values below about 7·0 (produced by high CO_2 tensions) but not with pH values as high as 8·96.

This paper describes more fully the effect of temperature and other factors on the development and hatching of herring. It forms part of a series of experiments (so far unsuccessful) to rear herring under controlled conditions to a stage where the vertebrae can be counted, and to see whether these counts can be correlated with differences in environmental conditions such as temperature.

During the experiments, preliminary data have been obtained, some of which are included here. Further details of the effect of temperature on the number of myotomes in young larvae will be published separately (Part III, Mar. Res. Scot., 1956).

METHODS

General

Experiments were carried out over a period of three years commencing in February 1953 and continuing to the spring of 1956. There are two groups of herring, one spawning in the autumn and the other in the spring and seven separate experiments were carried out, i.e. in the spring and autumn of 1953, 1954 and 1955 and the spring of 1956. Gonads were obtained from spring spawning fish in the Clyde and the North Minch and from autumn spawning fish off the Scottish east coast. Hatching failed in spring 1953 and no details are given of these experiments. Most of the work refers to data collected in autumn 1954 and spring 1955.

The gonads were removed from ripe fish and stored dry in sterile jars at 4°C for transport to the laboratory (see Blaxter, 1955).

Most of the experiments concerned the direct crosses SxS and AxA but it has also been found possible to store herring spermatozoa for six months (Blaxter 1955) and this enabled experiments to be made on the development of "hybrid" larvae produced from parents of different spawning groups.

Apparatus

Herring eggs normally stick to the substratum. On arrival at the laboratory the eggs were usually fertilised on ground glass plates but other substrata were also used. They were fertilised in sea water at the same temperature as the water in which they were later incubated. After about five minutes they were transferred to the incubation tanks.

The tanks used were accumulator jars, painted black, measuring 52 × 28 × 41 cm and containing about 50 litres of sea water. They were placed on thick cork sheets resting on sponge rubber and the walls were insulated with cork sheeting or glass wool "mattresses".

Each tank was adjusted to a different temperature which was kept as constant as possible throughout the experiments. Temperature was controlled by mercury-toluene thermostats which operated, through relays, either heating elements or centrifugal pumps circulating cooled glycerol and water through glass coils immersed in the tanks.

Each tank was divided into three compartments by muslin screens. The centre one contained either the heating element or cooling coil, a glass stirring rod operated by a motor above the tank, and an airlift filtering and aerating system. This raised water from the bottom of the tank, passed it through cotton wool and over marble chips at a rate of about 500 cc per minute, and then returned it to the surface of the two end compartments. These compartments contained perspex supports for the glass plates used as substrata. One compartment contained the thermostat bulb.

Environmental Conditions

(1) Water. This was coastal water obtained about six miles north-east of Aberdeen and stored in carboys for about two weeks before use. It was not changed during the course of an experiment.

After the first experiment it was decided to sterilise the water as the eggs became infected by hypotrichous ciliates. Sterilisation was usually carried out by heating the water twice to 70°C and then filtering through Doulton filter candles but on one occasion a chlorination method was used. The available chlorine in sodium hypochlorite was determined by titration with sodium thiosulphate. Experiments on the ciliates showed that a solution of ten parts per million was lethal. The amount of sodium hypochlorite calculated to give 10 p.p.m. was added to the tanks and after thirty minutes the required amount of sodium thiosulphate was added to neutralise excess chlorine. Orthotoluidine was used as an indicator of free chlorine and excess chloride was reduced by adding distilled water.

TABLE I

Details of Temperatures

Date	Tank	Designed Temperature (°C)	Observed Mean Temperature During Incubation	Standard Deviation of Reading
Autumn 1953	(1)	14·5	14·23	±0·370
	(2)	11·5	11·43	±0·230
	(3)	8·5	8·45	±0·159
Spring 1954	(1)	14·5	14·68	±0·168
	(2)	11·5	11·54	±0·158
	(3)	8·5	8·82	±0·270
	(4)	5·5	5·27	±0·211
Autumn 1954	(1)	14·5	14·51	±0·269
	(2)	11·5	11·31	±0·344
	(3)	8·5	8·66	±0·233
	(4)	5·5	5·68	±0·697*
Spring 1955	(1)	14·5	14·48	±0·127
	(2)	11·5	11·01	±0·714†
	(3)	8·5	8·43	±0·326
	(4)	5·5	5·10	±0·266
Autumn 1955	(1)	18·0	17·97	±0·288

*becomes ± 0·354 if one high reading omitted. †becomes ±0·487 if one low reading omitted.

(2) Temperature. Three tanks were used initially with temperatures set at about 8·5°C, 11·5°C and 14·5°C, and later a fourth tank set at 5·5°C was added. This range of temperature included the average temperatures at which spring and autumn spawners develop. The temperatures were normally taken twice per day and usually kept within reasonable limits. Mean temperatures during entire incubation periods are given in Table I with the standard deviation of the individual readings. Occasionally large variations occurred because of failures in some part of the apparatus but these were immediately corrected.

(3) Salinity. Salinity was determined at intervals of four or five days by titration against silver nitrate. Distilled water was added to compensate for evaporation when necessary.

Table II shows the mean salinity in the tanks for each experiment. The trend towards higher salinities at higher temperatures is to some extent exaggerated as a high reading was immediately corrected by adding distilled water.

TABLE II
Details of Salinities

Date \ Designed Temp. (°C)	5·5	8·5	11·5	14·5
Autumn 1953 ..	—	34·7	34·9	35·2
Spring 1954 ..	34·5	34·9	35·2	34·9
Autumn 1954 ..	34·3	34·3	34·7	34·8
Spring 1955 ..	34·2	34·4	34·6	34·7
Mean	34·3	34·6	34·9	34·9

(4) Oxygen. Measurements were made at intervals by the Winkler method. It was found impossible to keep the absolute value of dissolved oxygen the same in each tank due to temperature differences which would have required supersaturation at higher temperatures. The percentage saturation at the different temperatures, however, varies only slightly and Table III shows the average value of oxygen tension and percentage saturation over the range of experiments. This order of percentage saturation is very similar to that found in the sea at the same temperatures in the region of the spawning grounds.

TABLE III
Details of Dissolved Oxygen

Designed Temperature (°C)	Average Oxygen Tension (c.c/l.)	Saturation Tension (c.c/l.)	% Saturation
5·5	6·85	7·09	96·6
8·5	6·35	6·64	95·6
11·5	5·85	6·25	93·6
14·5	5·48	5·92	92·6

Care was taken by using indirect aeration to keep the oxygen tension below full saturation. No instance of gas disease (see Henly, 1952), was noticed.

(5) Light. The experiments were carried out in a darkened room but some light entered during the day, affecting the tanks equally. The maximum brightness measured on an Eel photometer was about 3 candles/sq. ft.

An experiment was later carried out to assess the effect of light on hatching (page 17).

(6) pH value. This was determined calorimetrically and was maintained between 7·5 and 8·0, any acidity being buffered by marble chips present in the filtering system.

Treatment of Material

The eyes of live embryos showed up clearly under a bright light so that it was easy to observe the extent and distribution of prospective hatching by shining a light on the glass plates shortly before hatching was expected.

Samples of larvae were anaesthetised in 2% magnesium chloride or 1% urethane and fixed in 4% formol-saline. The glass plates were kept in 4% formol-saline for analysis of distribution and percentage hatching of the eggs.

The time to 50% hatching and 50% yolk sac absorption was determined as near as possible in each experiment. This criterion, rather than the time to first instance of hatching or yolk sac absorption, was adopted.

RESULTS

The Effect of Egg Density on Development

In the experiments of autumn 1953 very few eggs hatched. An analysis of the distribution of hatched eggs showed that, where eggs were closely packed together in a thin layer on the surface of glass plates, only those at or near the periphery reached the eyed or hatching stage.

The percentage of eggs reaching the eyed or hatching stage can be related to the length of the periphery of the egg mass. This is shown in Table IV (for autumn 1953).

TABLE IV

Survival of Eggs (Autumn, 1953)

Number of Eggs	Peripheral Length of Egg Mass (cm)	Peripheral Length per Egg (μ)	% Eyed	% Hatched	Designed Incubation Temperature (°C)
3000	31	103·0	6	3	11·5
2700	52	193·0	16	9	8·5
2700	54	200·0	11	9	11·5
1800	40	222·0	13	9	14·5
2100	56	267·0	18	13	8·5
1900	77	405·0	30	17	8·5
900	61	678·0	26	25	14·5
600	44	730·0	37	33	11·5

In Fig. 1 the peripheral length per egg is plotted against percentage hatching, irrespective of temperature which showed no significant effect on development on this occasion.

The apparent linear relationship between percentage hatching and peripheral length per egg can be analysed further. A regression analysis gives $r=0{\cdot}98$ ($P=0{\cdot}001$). The equation for the regression line is—

$$Y=5{\cdot}0+23{\cdot}4X \quad \ldots\ldots\ldots\ldots 1$$

where X is percentage hatching and Y peripheral length per egg in μ.

This means that in a large laminar egg mass a high percentage hatching will only be obtained if the eggs are well spaced out. In a small laminar egg mass close grouping of the eggs will be less serious.

Other effects of crowding were observed when eggs were grouped more than one layer thick on glass plates. In this case only the outside ones developed and hatched, though the inner ones were fertilised. In experiments subsequent to autumn 1953 eggs were always well spread out over glass plates and this usually led to a higher percentage hatching.

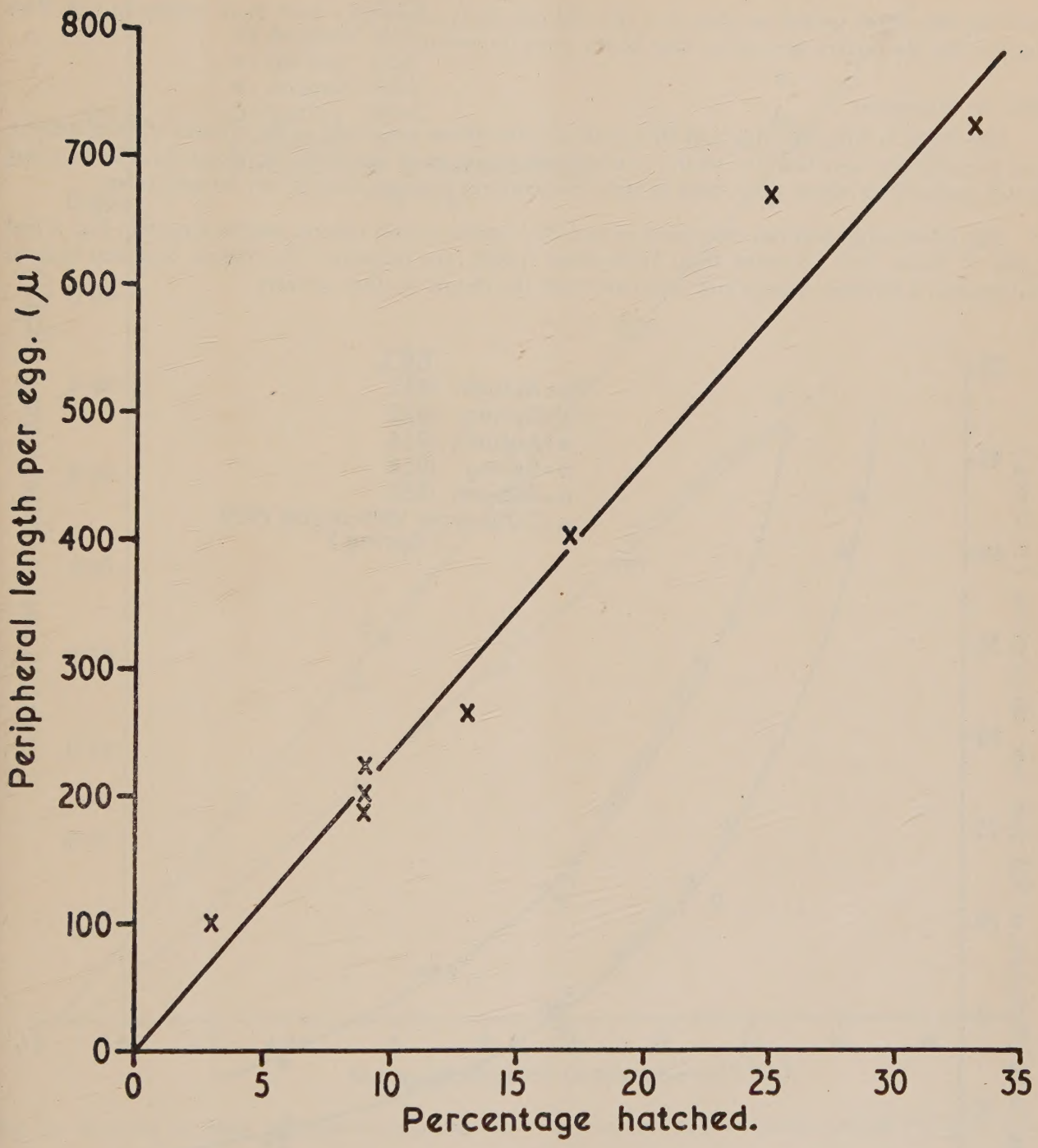

Fig. 1 Showing the effect of peripheral length of an egg mass on percentage hatching.

Lea (1930) and Runnstrøm (1941) found that herring spawn, naturally deposited in large clumps, gave a low percentage hatching. Considerable importance was attributed to this factor in its effect on brood survival. Ewart (1883) however showed that herring spawning in aquaria, deposited their eggs in small conical clumps. The reduction in percentage hatching may be due to mechanical interference with development or to insufficient surface area for the exchange of respiratory and excretory materials. It was found in rearing experiments that eggs closely spread on nylon gauze gave a higher percentage

hatching than those spread on glass, but this did not apply when they were more widely spread. This suggests that the surface area factor may be the more important.

Time to Hatching

The times to 50% hatching and 50% yolk sac absorption were used as the criteria of development. Low temperatures were found to retard, and high temperatures to accelerate, development. The hatching period ranged from about three days at high temperatures to seven days at low temperatures.

The relationship between temperatures and the developmental criteria used is shown in Fig. 2 and Table V. Some data extracted from Williamson (1909), are included. The curves obtained may be examined in a number of ways and compared with the results of other workers.

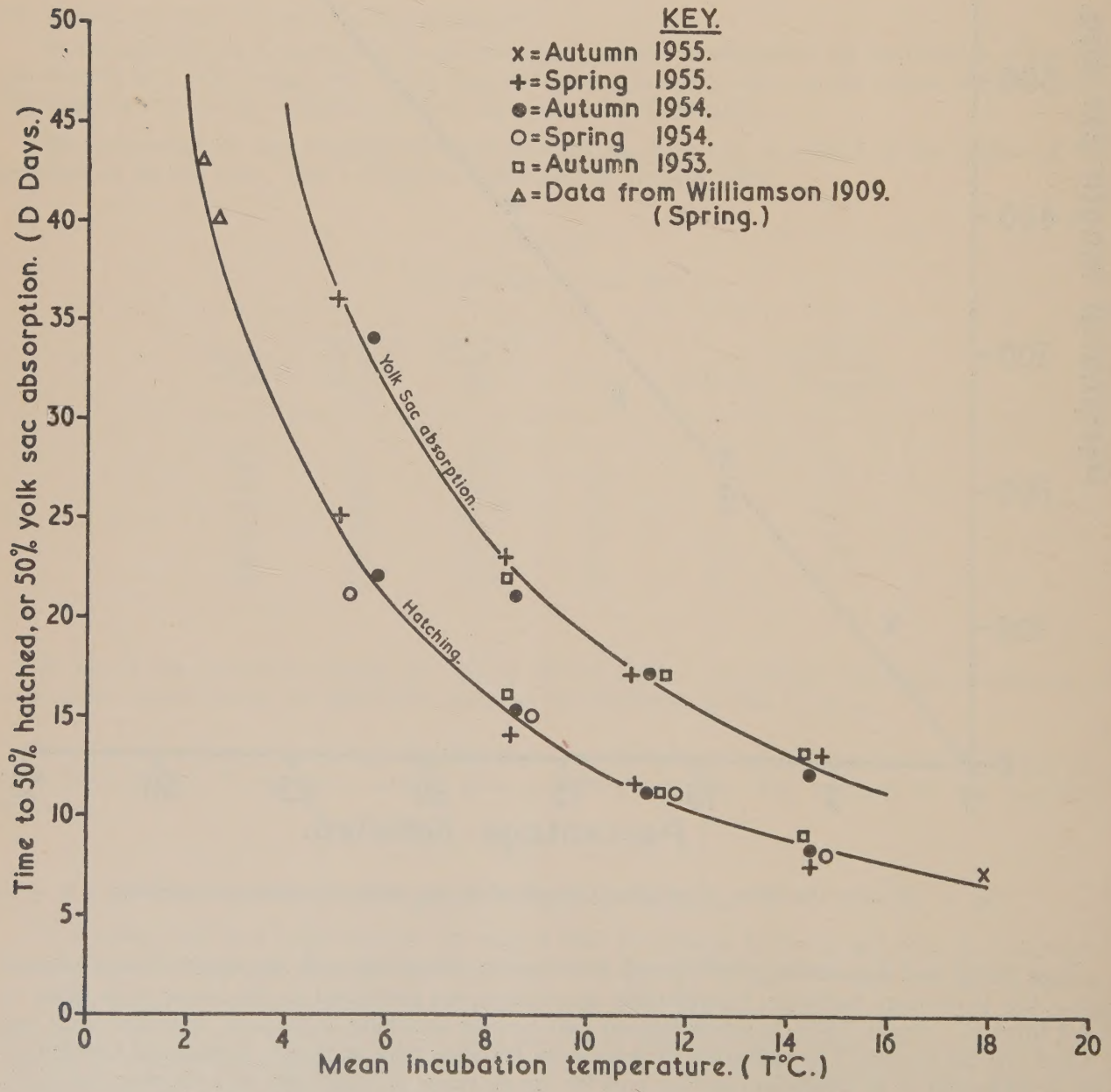

Fig. 2 Showing the effect of temperature on the time for development.

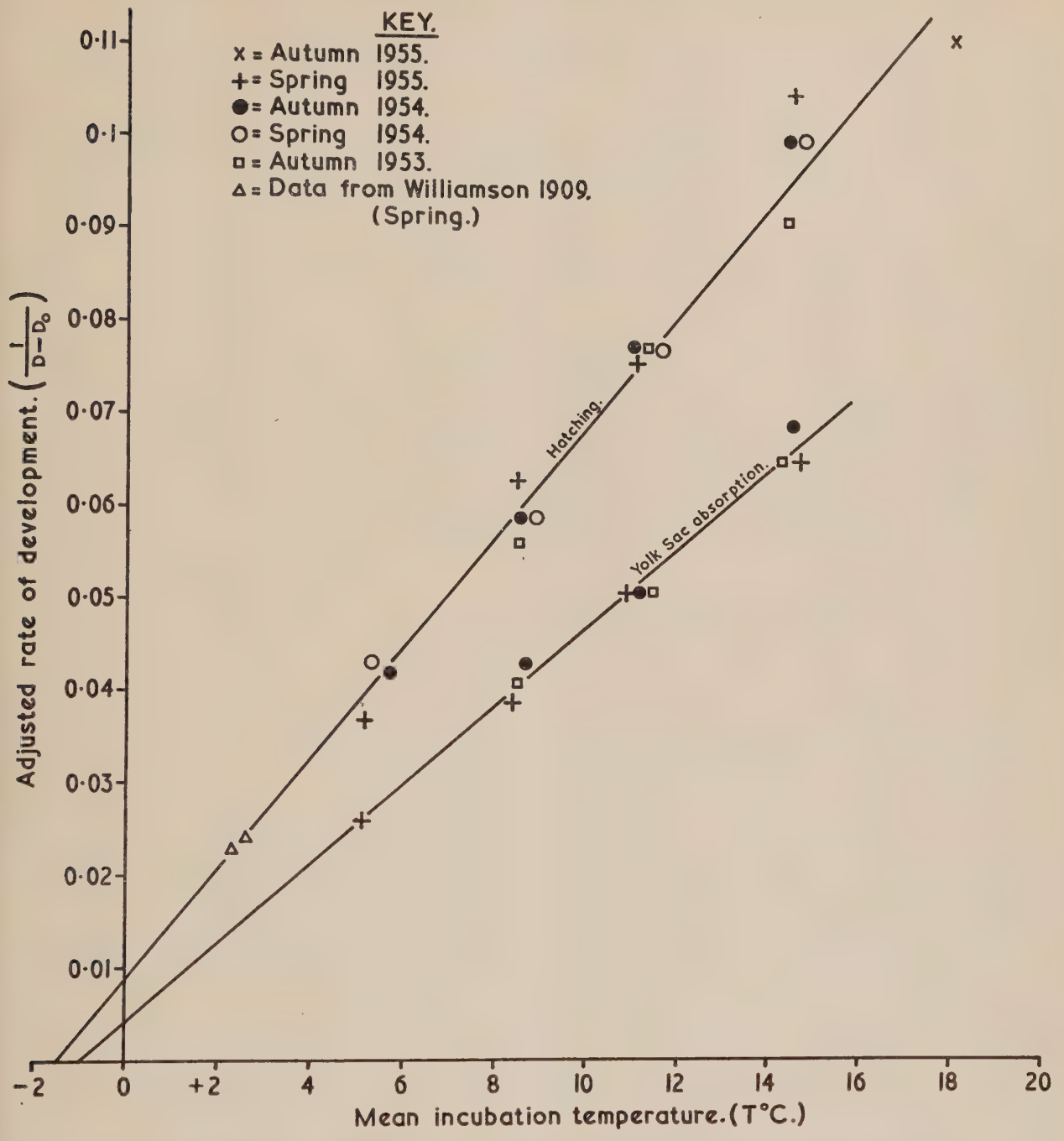

Fig. 3 Showing the effect of temperature on the rate of development and the position of the biological zero.

(*1*) *Estimation of biological zero or threshold temperature*

As the rate of development decreases with falling temperature, at some point (called the biological zero temperature) development will cease. In the same way, at a theoretically maximum temperature, the rate of development will be greatest.

The biological zero is used in estimating the amount of heat required by a developing organism to reach a given stage of development, or for calculating the length of time required by an organism to reach a given stage of development when incubated at different temperatures.

TABLE V

Development at Different Temperatures

Hatching				Yolk Sac Absorption			
Date	Mean Temperature T. (°C)	Days to 50% Hatching D (Days)	Rate $\frac{1}{D}$	Date	Mean Temperature T. (°C)	Days to 50% Absorption D (Days)	Rate $\frac{1}{D}$
Autumn 1955	17·97	7	0·1428	—	—	—	—
Spring 1954	14·68	8	0·1250	Autumn 1954	14·51	12	0·0833
Autumn 1954	14·53	8	0·1250	Spring 1955	14·48	13	0·0769
Spring 1955	14·53	7·5	0·1333	Autumn 1953	14·23	13	0·0769
Autumn 1953	14·35	9	0·1111	—	—	—	—
Spring 1954	11·54	11	0·0909	Autumn 1953	11·43	17	0·0588
Autumn 1953	11·45	11	0·0909	Autumn 1954	11·31	17	0·0588
Autumn 1954	11·28	11	0·0909	Spring 1955	11·01	17	0·0588
Spring 1955	11·13	11·5	0·08696	—	—	—	—
Spring 1954	8·82	15	0·0668	Autumn 1954	8·66	21	0·0476
Autumn 1954	8·66	15	0·0668	Autumn 1953	8·45	22	0·0455
Spring 1955	8·49	14	0·0714	Spring 1955	8·43	23	0·0435
Autumn 1953	8·46	16	0·0625	—	—	—	—
Autumn 1954	5·70	22	0·0455	Autumn 1954	5·68	34	0·0294
Spring 1954	5·27	21	0·0476	Spring 1955	5·10	36	0·0278
Spring 1955	5·15	25	0·0400	—	—	—	—
W	2·6	40	0·025	—	—	—	—
W	2·3	43	0·0233	—	—	—	—

W=data from Williamson (1909)

The data in Table V can be used to estimate the biological zero. The curves in Fig. 2 represent the best fit of these data and were obtained in the following way. The distribution of points suggested that the formula:—

$$(T-T_o)\,(D-D_o)=K \quad \text{or} \quad T=T_o+\frac{K}{D-D_o} \qquad \ldots\ldots\ldots\ 2$$

would provide the best fit, where D is the time taken to reach a given stage of development at temperature T, K is a constant, T_o the temperature at which development takes infinite days (the biological zero temperature) and D_o the time to hatching at infinite temperature (theoretical only).

It was considered that a satisfactory procedure for calculating the values in the equation, and therefore the biological zero, was as follows. The best fit for the two sets of points was first drawn by eye. Arbitrary points on these curves were taken and graphs drawn relating $\frac{1}{D}$ against T and $\frac{1}{T}$ against D. Two curvilinear regressions resulted for each set of points from which rough values of T_o and D_o (i.e. T_x and D_x) were obtained by extrapolation. Using these estimates of T_o and D_o, new graphs were drawn of $\frac{1}{D-D_x}$ against T and $\frac{1}{T-T_x}$ against D. Linear plots were obtained which indicated that equation 2 was of the right form and that satisfactory estimates of T_o and D_o had been made.

A regression of $\frac{1}{D-D_o}$ against T was then drawn using the estimated value of D_o (i.e. D_x) and the experimental values of D and T. This is shown in Fig. 3, the line of best fit and T_o being obtained by linear regression analysis. The slope of the line gives K in equation 2 so that all values for the equations are available. These are shown in Table VI. A linear regression analysis could not have been carried out on the raw data as it would presuppose that $T=T_o+\frac{K}{D}$ i.e. that $D_o = o$.

TABLE VI

Value of Biological Zero

Stage	Estimated Value of D_o (Days) i.e. (D_x)	Calculated Value of T_o (Biological Zero) (°C)	Calculated Value of K	Equation 2
To Hatching	−2·00	−1·34	165·0	(T+1·34) (D+2·0)=165·0
To Yolk Sac Absorption	−2·50	−0·77	231·0	(T+0·77) (D+2·5)=231·0

These calculations are made with the proviso that extrapolation from regressions may be a dangerous practice without knowledge of the processes underlying the relationship. It is interesting to note that the values obtained for the biological zero differ when hatching and yolk sac absorption are taken as the criteria. This may indicate the limitation of the analysis. The danger of extrapolation is obvious, for the organism would have died within a few degrees of the upper limit of temperature in the experiments but extrapolation is the only method of obtaining a value for the biological zero without carrying out tedious experiments at low temperatures.

Soleim (1942) working on the Norwegian herring estimated the biological zero at −1·2°C for hatching. The temperature during incubation was rather variable but a linear relationship was obtained by plotting $\frac{100}{D}$ against T.

Johansen and Krogh (1914), analysing their own and Dannevig's (1895) data, assumed that plots of $\frac{1}{D}$ against T for the development of teleost embryos were linear. Some of their regressions, however, might well be curvilinear, giving different values for the biological zero.

The work of Reibisch (1902) showed that for practical purposes the product of temperature and time to a given stage of development in teleost embryos was constant for a particular species, the value of the temperature being taken from the biological zero. This could be expressed as—

$$(t_1+x)n_1=(t_2+x)n_2 \quad \ldots\ldots\ldots\ldots \quad 3$$

where n_1 and n_2 are the days taken to reach a given stage of development at temperatures t_1 and t_2, x being the biological zero.

Dannevig (1948) used this method for herring, assuming the biological zero was O°C, and found that the product varied widely. He grouped the products into four and suggested that each group was racially distinct. The disadvantage of Reibisch's method for estimating the biological zero is apparent in that only two empirical observations can be used while by analysis of a greater range of data the best value for the whole range is found. If pairs of values for t_1, t_2, n_1 and n_2 are taken from Table V and substituted in equation 3 the estimates of x will be found to vary considerably.

(2) *Effect of temperature on rate of development*

The rate of chemical reaction is increased by rise in temperature. Van't Hoff's law states that "for every rise of 10°C the velocity of a reaction is doubled or trebled". His concept of the temperature

coefficient, Q_{10}, has been applied to biological processes (see Barnes, 1937) and many of these have been found to follow Van't Hoff's rule. It has also been applied to fish development with the same result (see review by Battle, 1930).

The Arrhenius relationship introduces μ, the temperature characteristic or critical thermal increment. This concept has also been applied to biological processes (see Heilbrunn, 1943) and its use may be preferable to Q_{10} as it has a sounder theoretical basis. Ahlstrom (1943) found an average value of 22,500 for development of the Pacific pilchard egg.

Values of temperature and time of development were taken from the curves in Fig. 2. In Table VII the values of Q_{10} and μ are given for 3 deg. C intervals between 3°C and 18°C for hatching and between 6°C and 15°C for development to yolk sac absorption. It appears that the variation in Q_{10} and μ over this relatively narrow range is far greater than that found in either chemical reactions or in many other biological processes. A decrease in values of Q_{10} and μ with rising temperature does occur in chemical reactions but it is spread over a much greater temperature range.

The conclusion is reached that while values of Q_{10} and μ might be used in certain circumstances to relate the development of the herring to other biological processes, development as judged by the criteria of hatching and yolk sac absorption does not follow the Arrhenius and Van't Hoff concepts except near the middle of the range of temperature used.

TABLE VII

Values of Q_{10} and μ

	Hatching						Yolk Sac Absorption			
Incubation Temperature T°C	3	6	9	12	15	18	6	9	12	15
Time in Days (D)	36	20·5	14	10·5	8	6·5	31·2	21	15·5	13
Rate of Development $K=\frac{1}{D}$	0·0278	0·0488	0·0714	0·0952	0·1250	0·1539	0·0321	0·0476	0·0645	0·0769
Q_{10}		6·5	3·6	2·6	2·5	2·0		3·7	2·8	1·8
μ		28100	20000	15500	14760	11500		20800	16400	9500

Johansen and Krogh (1914) and Battle (1930) found a similar result in the development of other fish. As valid Van't Hoff and Arrhenius relationships are found in many other biological processes, it seems that these results are exceptions; this may be explained by the range of temperature used. Thus the Q_{10} seems to be nearest its usual value (between 2·0 and 3·0) when the temperature is about that of normal development. At the extremes of temperature some factor upsetting normal development may be operating and this will alter the temperature coefficients.

The Effect of Temperature on Survival

The percentage of eggs fertilised and reaching the eyed and hatching stage at different temperatures and with different races is shown in Table VIII and Fig. 4. The numbers in italics refer to eggs incubated on nylon gauze while all others refer to eggs incubated on glass. Weighted means are given for each group of results.

TABLE VIII

Survival at Different Temperatures

Designed Incubation Temperature (°C)	Autumn 1954			Spring 1955		
	% Fertilisation	% Eyed	% Hatched	% Fertilisation	% Eyed	% Hatched
14·5	100 89 *100* *100*	77 30 } 51 *80* *44* } *62*	41 20 } 30 *38* *38* } *38*	99 99 99 100	8 29 } 17 23 17	6 17 } 11 17 12
11·5	100 94 *98* *100*	56 61 } 59 *57* *86* } *61*	22 37 } 30 *46* *67* } *55*	99 99 100 100	72 54 } 64 79 60	46 38 } 47 63 46
8·5	99 94 *100* *99*	53 60 } 56 *85* *56* } *69*	27 36 } 32 *19* *22* } *20*	99 100 100 100	77 81 } 83 82 90	47 40 } 59 65 72
5·5	99 85 *99* *99*	80 79 } 80 *86* *18* } *52*	36 29 } 32 *13* *9* } *11*	100 99 99 99	92 87 } 81 75 72	73 64 } 66 67 64

There is little consistent effect on percentage fertilisation of either temperature or race of herring. In the data of autumn 1954, there is no evidence of any significant effect of temperature or type of substratum in the eyed stage. On hatched eggs there is no significant temperature effect when incubated on glass, but there is when incubated on gauze; at low temperatures the percentage hatched is significantly lower ($P<0{\cdot}001$).

In spring 1955 the eggs were incubated only on glass. The differences in percentage eyed and hatched at different temperatures are highly significant ($P<0{\cdot}001$ for both sets of data). This is mainly due to the poor survival at high temperatures.

There is evidence here of a possible genetical difference between spring and autumn spawned herring. The autumn spawned fish show little effect of temperature on survival while the spring spawned fish show reduced viability when incubated at high temperatures. It may be assumed that autumn spawning in the sea takes place at about 9°C–14°C and spring spawning at about 5°C–8°C. If it were possible for herring to change the time of spawning during their lifetime the "spring" spawner might be at a disadvantage spawning in autumn temperatures though the converse would not be true. The above effect, however, might be due to acclimatisation temperature.

To ensure that percentage fertilisation was not connected with any particular part of the gonads, a simple experiment was carried out taking eggs and spermatozoa from either the anterior, middle or posterior regions of the gonads and fertilising them in different combinations.

There was no consistent reduced percentage fertilisation in any part of the gonads.

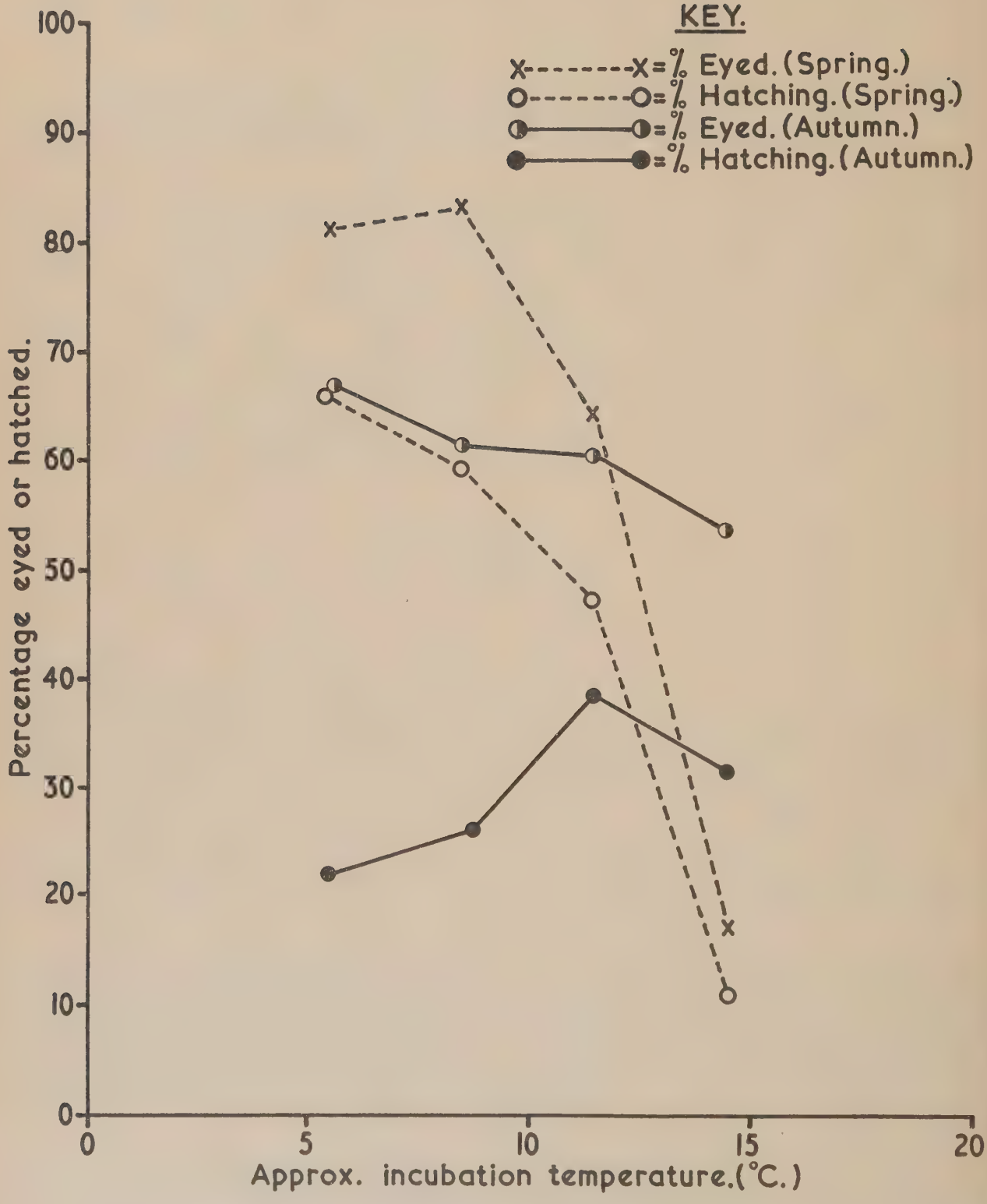

Fig. 4 Showing the survival of eggs of different spawning groups when incubated at different temperatures.

The Effect of Temperature on Size, Weight and Stage of Development

On hatching, the average herring larva has a large yolk sac. The lower jaw is just visible and pigmentation is diffuse. During subsequent growth the yolk is absorbed and the lower jaw grows forward. At the stage when the yolk has been completely absorbed the anterior margins of the upper and lower jaw are level. It is unlikely that the jaw is movable at this stage; the larva engulfs its prey by a darting movement.

The diffuse pigment gradually becomes more concentrated into spots along the gut. Pigment then appears above the anus between the gut and the body wall and small spots appear in the extreme caudal region.

(1) *Length*

There is a considerable increase in length and in the number of visible myotomes as the larva grows. Measurements at different temperatures, at different times of year and at different stages of development are shown in Table IX. The length given is the average for 10 larvæ and each pair of observations refers to the progeny of different crosses but with one parent common to both.

TABLE IX

Size of Larvae at Different Temperatures

	Autumn 1954				Spring 1955				Autumn 1955			
									Aut. × Aut.		Spr. × Aut.	
									♂	♀	♂	♀
Designed Incubation Temperature (°C) ..	14·5	11·5	8·5	5·5	14·5	11·8	8·5	5·5	11·5		11·5	
Average Length on Hatching (mm)	6·00 6·30	6·00 6·40	5·33 6·33	5·40 5·18	6·68 6·95	6·63 6·75	6·65 6·75	7·08 6·93	5·80		5·93	
Average Length on Yolk Sac Absorption (mm)	7·45 –	– –	7·35 –	– –	8·44 8·39	8·33 –	– –	8·16 –	–		–	

Data are also given for average length of larvae from the cross fertilisation of a spring male and an autumn female compared with the length when the same autumn female is crossed with an autumn male.

The length of larvae at hatching seems to decrease with lowering temperature in the autumn spawners and increase in the spring spawners. The former is in fact a spurious effect. The larvae incubated during autumn 1954 at 8·5°C and 5·5°C hatched unusually early and in three cases where this occurred the mean length at hatching was less than 6 mm. If this effect is ignored there appears to be little difference in length at different temperatures. On the other hand there is a very noticeable effect in the mean length of progeny of spring and autumn spawners. The overall mean of autumn spawned larvae at hatching was 5·86 mm and of spring 6·80 mm. The difference in mean is 0·94 mm and has a standard error of $\pm 0{\cdot}189$ which is significant ($P < 0{\cdot}05$). This result, however, is biased by the abnormally low values mentioned above.

The results obtained point to difference in mean length at hatching between spring and autumn spawners. A similar difference was shown by Schnakenbeck (1929) in the field. It is of importance that such measurements should be made at the same stage of development and before feeding commences. The difference may be of genetical origin, which would indicate a difference in the genotype of spring and autumn spawners, but on the other hand it might be due to difference in the size of the egg of spring and autumn spawners as a result of difference in feeding conditions for the adults prior to spawning. If

the latter were true one might expect that autumn spawners would have larger eggs and larvæ but this is not so. The data on the cross fertilisation support the view that the size of the larvae depends on the race of the mother, for the cross between a spring male and an autumn female yield larvae conforming more to the autumn average length. It may be that the size of larvae is then determined by the size of the egg, the autumn eggs being smaller, but whether this is a genetical effect or one produced by other conditions cannot be decided. Farran (1938) has shown that the eggs of spring spawning Irish herring are larger than those of autumn spawning fish and Fridriksson and Timmermann (1951) have described differences in the size of eggs of different herring races, as reported by a number of authors.

The control of conditions in this experiment is not completely satisfactory but as far as could be determined there was no factor which differed between the experiments in the spring and the autumn. The data from the other experiments in autumn 1953 and spring 1954 confirm the results shown in Table IX.

(2) Weight

The work of Gray (1928) and Merriman (1935) showed that salmonid fry were heavier at low incubation temperatures than at high ones. Weight data for herring larvæ are given in Table X. The average wet weight per larva in milligrams is given on hatching at different temperatures and the pairs of observations again refer to different crosses with one parent common to both.

TABLE X

Weight of Larvae at Different Temperatures

	Autumn 1954				Spring 1955				Autumn 1955			
									Aut. × Aut.		Spr. × Aut.	
									♂	♀	♂	♀
Designed Incubation Temperature (°C) ..	14·5	11·5	8·5	5·5	14·5	11·5	8·5	5·5	11·5		11·5	
Average Weight on Hatching (mg)	0·8 0·8	0·6 0·7	0·7 0·8	0·7 0·7	1·1 1·1	1·3 1·3	1·4 1·4	1·5 1·3	0·8 —		0·7 —	

A difference in average weight comparable with the difference in average length is found between the spring and autumn spawned larvae. The overall autumn mean is 0·73 mg, and the spring 1·30 mg. The difference in means, 0·57 mg, has a standard error of $\pm 0{\cdot}058$ which is highly significant ($P < 0{\cdot}001$). This may again indicate a difference in the genotype of the parents and as the spring × autumn cross has an autumn average weight the difference seems to be due to a maternal influence, probably egg size.

There is a tendency for the average weight to increase at low temperatures in the spring spawned larvae but the overall increase in average weight at low temperature is not significant ($F = 1{\cdot}04$, d.f. 3 and 9).

(3) Other characters

An examination of morphological characters such as position of the lower jaw, presence of pigment and size of yolk sac show that nearly always there is no difference in stage of development at hatching in larvae from different spawning groups or from different incubation temperatures. The exceptional group (of autumn 1954) was clearly hatched at an earlier stage of development than the others; the lower jaw was absent, pigment had not appeared and myotomes could not be distinguished beyond the anus.

The Effect of Light, Sterility and Water Movement

Developing eggs were incubated in four tanks at similar temperatures. Two tanks contained sterilised, and two unsterilised, sea water of the same origin. One tank containing each type of water was illuminated by a 60 W bulb giving an intensity of illumination of about 60 ft. candles on the glass plates acting as substrata. The other two tanks were kept in darkness. Each tank contained two glass plates over one of which a current of water was allowed to flow by means of an airlift system. The percentages of eggs reaching the eyed stage and hatching are given in Table XI with total numbers of fertilised eggs used in brackets. In all cases percentage fertilisation was between 98% and 100%.

TABLE XI

The Effect of Light, Sterility and Aeration on Hatching

		Lighted Sterilised	Lighted Unsterilised	Unlighted Sterilised	Unlighted Unsterilised
% Eyed	Ventilated	91·5 (967)	77·1 (1204)	88·5 (914)	85·6 (973)
	Unventilated	73·3 (1381)	88·8 (984)	88·7 (966)	87·7 (684)
% Hatched	Ventilated	30·0 (967)	35·5 (1204)	51·6 (914)	35·3 (973)
	Unventilated	23·5 (1381)	30·7 (984)	44·2 (966)	44·4 (684)

There appears to be no consistent difference in development due to sterility of the water or to the use of a current ventilating the eggs. There is, however, a significant difference in the percentage hatched in lighted and unlighted tanks. The combined percentages are as follows:—

Combined percentage hatched in sterilised water =37·3
" " " " unsterilised water =36·5

Combined percentage hatched on ventilated plates =38·1
" " " " unventilated plates =35·7

Combined percentage hatched in lighted tanks =29·9
" " " " unlighted tanks =43·9 ($P<0·05$).

CONCLUSIONS

The effect of temperature on the development of the herring does not seem to follow the pattern of many other biological processes. The Van't Hoff and Arrhenius relationships do not hold over the temperature range used, i.e. 3°C to 18°C, though they are most typical between about 6°C and 12°C. This indicates the susceptibility of development to temperature extremes outside the normal range.

Differences in the experimental results are found when the development of spring and autumn spawned herring is compared. The percentage hatching at different temperatures and length and weight at hatching are different in the two groups. This indicates that spring and autumn spawned herring have different characteristics. Whether these characteristics can be attributed to a difference in genetical constitution or to some factor, such as egg size, which is a result of environmental conditions at the time of spawning, is not known, but the evidence supports the former rather than the latter view.

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SUMMARY

Eggs from autumn and spring spawning herring have been artificially fertilised and incubated at different temperatures from 5°C to 18°C.

Developing eggs give a low percentage hatching when crowded together.

The effect of temperature on development has been considered mathematically: the biological zero has been calculated as −1·34°C for hatching.

The Van't Hoff and Arrhenius relationships for the effect of temperature on biological processes do not hold over the whole range of temperature used.

Under experimental conditions the percentage hatching at different temperatures and the length and weight of larvae on hatching of spring spawned herring differ significantly from autumn spawned herring. It is suggested that this may indicate a genetical difference between the spawning groups.

A higher percentage hatching was obtained in darkness than in light. No increase in percentage hatching was obtained by the use of sterilised water or by ventilating the eggs with currents of aerated water.

REFERENCES

Ahlstrom, E. H. 1943. 'Studies on the Pacific pilchard or sardine.' Spec. sci. Rep. U.S. Fish Wildl. Serv., no. 23.

Barnes, T. C. 1937. *Textbook of General Physiology*, pp. xxii+554. Philadelphia: Blakiston.

Battle, H. I. 1930. 'Effects of extreme temperatures and salinities on the development of *Enchelyopus cimbrius* L.' Contr. Canad. Biol., **5**, 107.

Blaxter, J. H. S. 1955. 'Herring rearing—I. The storage of herring gametes.' Mar. Res. Scot., 1955, no. 3.

Brook, G. 1885. 'On the development of the herring.' Rep. Fish. Bd Scot., **3**, 32.

—— 1886. Rep Fish. Bd Scot., **4**, 31.

Dannevig, A. 1948. 'Rearing experiments at the Flodevigen sea fish hatchery, 1943-1946.' J. Cons. int. Explor. Mer, **15**, 277.

Dannevig, H. 1895. 'The influence of temperature on the development of the eggs of fishes.' Rep. Fish. Bd Scot., **13**, 147.

Ewart, J. C. 1883. 'Natural history of the herring.' Rep. Fish. Bd Scot., **2**, 61.

Farran, G. P. 1938. 'On the size and number of the ova of Irish herring.' J. Cons. int. Explor. Mer, **13**, 91.

Ford, E. 1929. 'On the artificial fertilisation and hatching of herring eggs under known conditions of salinity with some observations on the specific gravity of the larvæ.' J. Mar. biol. Ass. U.K., **16**, 43.

Fridriksson, A. and Timmerman, G. 1951. 'Some remarks on the eggs of herring (*Clupea harengus*, L.) . . .' J. Cons. int. Explor. Mer, **17**, 261.

Gray, J. 1928. 'The growth of fish: II and III.' J. exp. Biol., **6**, 110.

Hata, K. 1929. 'On the influence of the duration of the time of vibration upon the development of fish eggs.' J. Fish. Inst. Tokyo, **24**, 121.

—— 1949. 'Effect of light on hatching of fish eggs.' Bull. Jap. Soc. sci. Fish., **15**, 375.

Heilbrunn, L. V. 1943. *An Outline of General Physiology*, 2nd ed., pp. xii+748. Philadelphia: Saunders.

Henly, E. 1952. 'The influence of the gas content of sea water on fish and fish larvæ.' Rapp. Cons. Explor. Mer, **131**, 24.

Johansen, A. C. and Krogh, A. 1914. 'The influence of temperature and certain other factors upon the rate of development of the eggs of fishes.' Publ. Circ. Cons. Explor. Mer, no. 68, 1.

KAJIYAMA, E. 1929. 'On the influence of temperature and specific gravity upon the development of the eggs of *Pagrosomus major*.' J. Fish. Inst. Tokyo, **24**, 117.

KELLEY, A. 1946. 'Effect of abnormal CO_2 tension on the development of herring eggs.' J. Fish. Res. Bd Can., **6**, 435.

KUPFFER, C. 1878. 'Die Entwicklung des Herings im Ei.' Wiss. Meeresuntersuch., **3**, 175.

LEA, E. 1930. 'Mortality in the tribe of Norwegian herring.' Rapp. Cons. Explor. Mer, **65**, 100.

MERRIMAN, D. 1935. 'The effect of temperature on the development of the eggs of the cut throat trout (*Salmo clarkii clarkii*, Richardson).' J. exp. Biol., **12**, 297.

MEYER, H. A. 1878. 'Beobachtungen über das Wachsthum des Herings im westlichen Theile der Ostsee.' Wiss. Meeresuntersuch., **3**, 227.

NAKAI, N. 1927. 'On the influence of water temperature upon the development of the eggs of *Leuciscus hakensis*, Günther.' J. Fish. Inst. Tokyo, **22**, 73.

REIBISCH, J. 1902. 'Ueber den Einfluss der Temperatur auf die Entwicklung von Fisch-Eiern.' Wiss. Meeresuntersuch., **6**, 213.

RUNNSTRØM, S. 1941. 'Quantitative investigations on herring spawning and its yearly fluctuations at the west coast of Norway.' Fiskeridir. Skr. Havundersøk., **6** (8), 1.

RYDER, J. A. 1882. 'On the retardation of the development of the ova of the shad (*Alosa sapidissima*) with observations on the egg fungus and bacteria.' Bull. U.S. Fish. Comm., **1**, 177 and 422.

SENO, H., EBINA, K. and OKADA, T. 1926. 'Effect of temperature and salinity on the development of the ova of a marine fish *Calostomus japonicus*.' J. Fish. Inst. Tokyo, **21**, 73.

SCHNAKENBECK, W. 1929. 'Entwicklungsgeschichtliche und morphologische Untersuchungen am Hering.' Ber. dtsch. Komm. Meeresforsch., **5**, 19.

SOLEIM, P. A. 1942. 'Årsaker til rike og fattige årganger av sild.' Fiskeridir. Skr. Havundersøk., **7** (2), 1.

WILLIAMSON, H. C. 1909. 'Experiments to show the influence of cold in retarding the development of the eggs of the herring (*Clupea harengus* L.), the plaice (*Pleuronectes platessa* L.) and the haddock (*Gadus aeglefinus* L.).' Rep. Fish. Bd Scot., **27**, 100.

—— 1911. 'Experiments in retarding the development of eggs of the herring.' Rep. Fish. Bd Scot., **28**, 16.

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